Association between traditional Faroese diet and vitamin D levels measured in umbilical cord blood

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There are no conflicts of interest to declare in this study

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Abstract

Background/objectives: Populations of northern latitude like the Faroe Islands may depend on marine diet as the major dietary vitamin D source. Intake of traditional Faroese marine food has declined and a lower vitamin D concentration is observed in the latest Faroese birth cohort, Cohort 5 compared with a previous. Our aim was to examine whether changes in marine diet and/or other cohort differences were associated with the observed decrease in average cord 25-hydroxy vitamin D (s-25(OH)D) concentrations. Furthermore, because only scarce information on cholecalciferol content in Pilot Whale exists, analyses of vitamin D content in Pilot Whale was performed.

Subject/Methods: In this cross sectional study, data was obtained from two different birth cohorts. Data included pregnant women and their offspring with obstetrical data from 579 and 447, diet data from 267 and 433 and vitamin D content in cord s-25(OH)D from 267 and 433 individuals from respective Cohort 3 and Cohort 5. Furthermore, vitamin D supplement data from 357 pregnant women in Cohort 5 was available. In 33 Pilot Whale samples, the nutrient vitamin D content was examined.

Results: In a multivariable regression a higher body mass index, fewer summer-birth's and a declined percentage of prospective parent's higher educational level may explain parts of the changed cord s-25(OH)D.

Conclusion: The decreased intake of marine food was not associated to decreased vitamin D level in cord blood. Pilot Whale and Blubber does not appear to be a dominant source of Vitamin D for women in Cohort 3 and Cohort 5.

Introduction

The prevalence of vitamin D deficiency is high worldwide. It is estimated that over a billion people worldwide are affected (1). The main function of vitamin D is to maintain the calcium and phosphate homeostasis. It is well known that vitamin D deficiency can lead to osteoporosis and fracture in elderlies and rickets in children(1). However recent studies also suggest that vitamin D has so called non calcemic effects as vitamin D deficiency increase the risk of several autoimmune disease including type 1 diabetes, type 2 diabetes and cardiovascular disease (2-4) and depression (5).

Vitamin D from diet mainly comes from fatty fish such as salmon and halibut but also cod or haddock liver oil, seals and some whale species (6). Humans get most of their circulating vitamin D from exposure to UVB, sunlight. However, some epidemiological studies have found that higher latitude is associated with lower level of plasma vitamin D in the population (7) and that in north Greenland, traditional diet, including sea mammals and free-living fish, has an impact on the plasma vitamin D level (8). Furthermore, a study of vitamin D level in an elderly population in the Faroe Islands (62°N) found an association between diet, time of the year for blood sampling, BMI and plasma vitamin D level. In that study the vitamin D level (measured a 25-hydroxy vitamin D, 25(OH)D) was low, < 50% had vitamin D levels over 50 nmol/l (9), which is considered as sufficient (10).

Traditional Faroese food consists mainly of fish, seabirds, whale meat and blubber (11). It is known that the Faroese food richest on cholecalciferol, is cod liver oil, salmon, halibut, puffin, mackerel and herring(6), which supposedly should ensure a sufficient s-25(OH)D concentration during winter months, a period of minimal sun exposure. It has been a

general Faroese assumption that pilot whale meat and blubber were rich on cholecalciferol (12, 13), but this has never been studied.

However in the Faroe Islands the traditional food habits have changed during the last three decades. In the Faroese study of dietary habits in 1982 including 331 Faroese, older than 14 years, they reported a daily intake of 12 g meat and 7 g blubber (14), while a study in 2001 including 148 pregnant women, the participants reported eating about 1.5 g meat and 0.6 blubber (15). These changes may have had an influence on s-25(OH)D concentrations.

In 1977 the chief medical officer in the Faroe Islands announced that liver from pilot whale was polluted, and advised to stop using the liver and kidneys for human diet, and to reduce consumption of whale meat and blubber to once a week. In 1985 studies revealed that the Pilot Whale had unhealthy amounts of PCB, mercury and other neurotoxic substances (15). This led to recommendations on the restriction of Pilot Whale intake. This entailed that researchers started a project named Children's Health and the Environment in the Faroes (CHEF-project), with the aim to determine effects of exposures to marine toxicants on the human health in prospective birth cohort studies (www.chef-project.dk). Follow-up of these cohorts hereafter led to the most recent recommendation, anno 2012, to completely abstain from Pilot Whale consumption (16). Following these new recommendations, recent unpublished data has demonstrated a lower umbilical cord blood vitamin D concentration in Faroese Cohort 5 (2007- 2009) compared to Cohort 3 (1997-2000). We therefore examined the association between intake of marine food, inclusive Pilot Whale meat and blubber, in pregnant women and concentration of 25(OH)D in umbilical cord blood. Furthermore, we assessed whether differences in cohort variables

could explain the difference in average 25(OH)D concentrations. Finally, as information on cholecalciferol content in pilot whale is scarce, nutrient analyses were performed.

Materials and methods

Study design and participants

This cross-sectional study was conducted from two different Faroese birth cohorts from the Chef-project, Cohort 3 and Cohort 5. Pregnant women were invited to join the CHEF cohort study. Of those who accepted, relevant obstetrical data were collected and measurements of contaminants were performed on blood, umbilical cord, hair and milk samples. The children were then followed with numerous clinical studies, of especially the central nervous system and at the same time blood samples were taken. Mothers and children's diet were clarified by questionnaires.

The cohort 3 consists of 656 births in Tórshavn between November 1997 and March 2000 and was formed after Faroese health authorities had introduced a dietary recommendation to reduce intake of especially Pilot Whale meat and blubber to lower the exposure to methyl mercury. Cohort 5 is the most recent birth cohort and consists of 490 childbirths (between October 2007 and April 2009). Further information about the Chef project and more detailed descriptions are described elsewhere (www.chef-project.dk).

As showed in figure 1, all participants from Cohort 3 and Cohort 5 that had full information regarding umbilical cord s-25(OH)D and obstetrical and diet questionnaire data; such as the mothers age, body mass index (BMI), diabetic illness, date of childbirth, intake of fish, seabird, Pilot Whale meat and blubber were included.

For Cohort 3 570 observations on obstetrical data were available, 570 vitamin D measurements and 267 diet questionnaire data. For Cohort 5 447, 448 and 433

observations were available, respective. Furthermore, data on vitamin D supplement of was available in Cohort 5 from 357 participants. A total of 570 observations and 447 observations were available from Cohort 3 and Cohort 5, respective.

Measurement of umbilical cord blood 25(OH)D

In short, following storage at -80°C, the concentrations of serum 25(OH)D2+3 were analysed using liquid chromatography-tandem mass spectrometry (LC-MS/MS) on a TurboFlow column (Thermo Scientific San Jose, CA, USA), as previously described [28]. Vitamin D analysis were performed of both vitamin D2 and D3 and reported as total vitamin D (i.e., s-25(OH)D).

Measurement of pilot whale nutrient content of cholecalciferol

To obtain information on Pilot Whale content of cholecalciferol, 33 pilot whale samples of each 100 g, 15 meat and 18 blubber samples, were collected from four different herds during the summer 2015 in the Faroe Islands. The first 29 samples were kindly provided by The Environmental Agency who collects samples at every herd driven ashore, and provides them with information about sex, age, size and time of the year for killing; these samples were all taken from around the genital opening, a piece of the whale not usually used for consumption, because of its poor quality. Thus, decision was made to analyse one sample of meat and three of blubber collected for the purpose of eating and obtained from private households. All 33 samples were analysed at Eurofins, Vejen, Denmark by standard laboratory procedures, using analysis number A7252.

Statistical analysis

Descriptive data were presented as numbers (percent) or mean (standard deviation). To

identify significant differences in characteristics between our groups in Cohort 3 and 5, we used a t-test for continuous variables on: umbilical cord s-25OHD concentration, women's age, weight gain, BMI before pregnancy and BMI after weight gain. Chi Square test was used for categorical variables of the following groups: women's age, parity, BMI before pregnancy, diabetic illness, childbirths season, smoking, alcohol consumption, women's educational level and the highest education of the parents. The highest education of the parents refers to highest educational level of the women or her partner/father to be.
In regression models, 25(OH)D concentrations were examined both as a continuous variable as well as dichotomised at the level of 50 nmol/l, as concentrations below this level are considered 'inadequate' by the Institute of Medicine (10) and Danish authorities. In addition, Chi Square test was performed to assess correlation between marine diet as categorical yes/no and sufficient and insufficient vitamin D level between the two cohort groups. We tested following women's intake of marine food during pregnancy: Pilot Whale meat, pilot whale blubber, seabirds, fish at least twice a week and traditional marine diet habits. Seabird, Pilot Whale meat or blubber intake during pregnancy were reported as yes if the woman had eaten it at least once during pregnancy. Fish at least twice a week were reported as yes, if the women had eaten it at least twice a week during pregnancy. Traditional marine diet habits were defined as intake during pregnancy of all of the following: seabird, Pilot Whale meat and blubber, and fish at least twice a week (yes/no). To examine the association between 25(OH)D, possible predictors/covariates and intake of marine food, multivariable regression models were performed. Main model 1 (n = 1017) included the following a priori selected variables: parity, BMI before pregnancy, diabetic condition, childbirth-season, smoking, alcohol consumption, highest education of the parents and cohort identity. Due to a high number of missing regarding intake of marine

food, these were not included in the model 1 but performed in a second model 2 (n = 700) included all of the variables in model one as well as Pilot Whale meat and blubber, seabird and fish intake at least twice a week during pregnancy.

The distribution of model residuals were evaluated by visual inspection of residual plot and judged near normal.

Results were considered statistical significant when $P < 0.05$.

Statistical analyses were done using Stata version 14.0 (Stata Corporation, College Station, TX, USA).

Ethics

The Faroese Committee on Research Ethics has approved the cohort studies. The informed consents were given to the Department of Occupational Medicine and Public Health, Torshavn, Faroe Islands. The Head of the Department granted the authors access to the data.

Results

Baseline characteristics of the two cohorts are illustrated in table 1. Women in Cohort 5 were heavier, less smoking, higher and older than the women in Cohort 3. Furthermore, they had a higher BMI, higher weight gain during pregnancy, fewer births in the summer and fewer women had a long educational background.

Finally, umbilical cord serum 25(OH)D concentration was 2.6 nmol/l lower in Cohort 5 than Cohort 3 ($p < 0.05$).

Traditional marine diet

The dietary habits during pregnancy has changed during the two cohort periods, from Cohort 3 till Cohort 5 (table 2). In Cohort 3, 193 (72.3 %) of the women ate fish more than twice a week during pregnancy, whereas 252 (57.9 %) in cohort 5. In Cohort 3, 159 (59.5 %) eat Pilot Whale meat, 101 (37.8 %) Pilot Whale blubber and 171 (64 %) eat seabirds during pregnancy. For the women in Cohort 5 these intakes were respectively 88 (20.3 %), 25 (5.8 %) and 141 (32.6 %). Furthermore, 46 (17.2 %) women in Cohort 3 eat all of the above-mentioned traditional food items compared to 9 (2.1 %) in Cohort 5.

Predictors of umbilical cord s-25(OH)D concentrataion

In the multivariable regression model 1, BMI, gestational diabetes and multipara were significantly correlated to a lower s-25(OH)D concentration (all p-values >0.05). Whereas giving birth in summer season was correlated with higher s-25(OH)D concentration. In model 2 giving birth both in summer and autumn as well as one of the parents having a medium or long educational background was significantly correlated to a higher s-25(OH)D concentration. However, BMI, multipara as well as having gestational diabetes was significantly correlated to a lower s-25(OH)D concentration. Furthermore, the Cohort 5 group was found significantly correlated to a lower s-25(OH)D concentration. However not any of the marine food items were significantly correlated to s-25(OH)D.

Vitamin D analysis of Pilot Whale.

The vitamin D, cholecalciferol contents of Pilot Whale meat and blubber are shown in table 3. The mean cholecalciferol concentration in Pilot Whale meat was 0.63 µg/100g and pilot

blubber was 1.1 µg/100g although higher content was found in the edible parts compared with the samples collected from the genital opening.

Discussion

The traditional marine food habits have changed in the Faroe Islands from 1997 to 2009 (table 2), however changes in marine diet did not seem to influence the changes in s-25(OH)D level of the women in Cohort 5, tested by the dichotomous analysis and multivariable regression model (table 2 and 4). However, the women's general BMI seemed to explain a part of the decrease in s-25(OH)D level in Cohort 5. Furthermore, it seems that a lower number of summer births and decline in the educational level of the parents to be also may explain parts of the change in s-25(OH)D level among pregnant women in Cohort 5 (table 4). These associations are also demonstrated by other studies (19). However, there are still some unidentifiable period effects (from 1997 to 2009) of the difference between women in Cohort 3 and Cohort 5, that can not be explained, even taking into account BMI, summer births and higher education level (table 4). Furthermore, gestational diabetes also indicated a negative correlation to vitamin D concentration ($p < 0.05$). However, the diagnosis of gestational diabetes is different at the two periods and in both cohorts it may have been misclassified, hence results may be less valid. Finally, Pilot Whale meat and blubber does not appear to be a dominant source of Vitamin D for women in Cohort 3 and Cohort 5.

A similar study from the Faroe Islands regarding an elderly population found that consumption of Pilot Whale blubber increased the odds of having a serum 25(OH)D >80 nmol/l by a factor of 1.5 (9). Government recommendations for intake of Pilot Whale meat

and blubber have not been as strict for elderly as for pregnant women, until 2012, why a higher intake may explain the association to vitamin D in elderly.

The study of Andersen et al (18) found that vitamin D status was influenced by diet and season. Their study showed that vitamin D concentration increases by eating more traditional Inuit food items. However, there are at least two differences between the two population. Firstly, the villages included in the Greenlandic study are located further north of the Faroe Islands. Secondly, the Inuit diet consists of seal and larger whale types that have a higher cholecalciferol concentration (8) and therefore may supply a better vitamin D source.

Season of birth and cohort period effects

From analysing the data in Cohort 3, we found a discrepancy in vitamin D levels between being born in the spring in Cohort 3 and Cohort 5. There was a significant higher vitamin D level in Cohort 3 with a p-value of 0,0132.

Summer-births also had higher vitamin D levels in cohort 3 compared to cohort 5. Autumn had a small difference, with Cohort 5 a little bit higher and autumn were equal (figure 2). This cohort-effect can be due to lifestyle changes from the late nineties up to now.

Even after adjustment for differences in BMI, season of birth, parity and gestational diabetes, there are still differences between Cohort 3 and Cohort 5. Hence this difference must be based on unidentified factors not included in this model. One factor not included in this study is outdoor activity, and it is reasonable to think that there might have been some changes in outdoor activity

In the nineties it was common that jobs had an hour lunch break, and most people went home to eat between the hours of 12pm and 1pm, increasing the likelihood for sun

exposure in the middle of the day. In 1997, there were 12,091 cars in the Faroe Islands compared to 20,293 in 2009. According to research carried out by the Office of National Statistics (www.hagstovan.fo) the distances travelled by these cars were 133 and 232 million km per year respectively. This may indicate that people did spend more time outdoors in the period from 1997 than in 2009. There was no marked difference in the hours of sun in both periods. Statistics from www.dmi.dk show that sun-hours in the period of cohort 3 were on average 918 hours per year. There was a minimum of 777 hours and maximum 1042 hours. In cohort 5 there was on average 1018 hours of sunshine a year, with a minimum of 912 and maximum of 1139. This is the exact opposite of what we had expected since the vitamin D level is lower in the cohort 5 (www.DMI.dk). We have no data on outdoor activities.

According to the Norwegian study of Engelsen et al, it is not possible to synthesize D vitamin during the months October to February due to the Faroe Islands high latitude. However, for the rest of the year it should be possible to get sufficient vitamin D synthesis from the sun (7) when the weather allows it.

Our observations indicate the challenges that lie ahead for pregnant women who aim to maintain adequate 25(OH)D levels by simply relying on eating traditional Faroese food. In the study of the elderly in the Faroe Islands, Dalgård et al concluded that higher supplements of Vitamin D and/or fortification might be necessary to maintain adequate 25(OH)D levels in order to prevent diseases and to ensure optimal body function and bone health. Regarding the fact the majority of women in Cohort 5 report taking vitamin D supplements, and still do not reach a sufficient level, our recommendation is to increase

the recommended doses of vitamin D supplements to Faroese pregnant women from, as it is, 10 µg to 15 µg.

Pilot whale analysis

In order to analyse the data of traditional Faroese food, such as whale meat and blubber anticipated to be rich in vitamin D, more information about their content of cholecalciferol was needed. The Faroese Food- and Veterinary Authority has a food composition table over Faroese food, but no information of cholicalciferol in whale meat and blubber(6). However, our present analyses demonstrated that Pilot Whale blubber has a mean cholecalciferol content of 1µg/100g and Pilot Whale meat has mean of 0.53µg /100 g. According to www.hsf.fo this does not give pilot whale a place among the richest sources in traditional Faroese food.

The Canadian study of Kuhnlein et al (19) has measured levels of vitamin D in different kinds of seafood. They found higher levels of vitamin D in boiled blubber compared to raw. Although these samples were taken from different types of whales, it would be of interest to see if the same case is present for Pilot Whale blubber, especially due to the fact that the trend for consuming blubber has changed over the years from eating boiled blubber with boiled meat to eating the blubber salted and dried. However, our analyses do not suggest that whale meat and blubber have high content of vitamin D.

Strengths and limitations

Respectively 60% and 73% of the eligible population took part in each of the two cohorts, making it representable as a general population of pregnant women in Faroe Islands.

Our data was comparable between the two cohorts, even though they were collected for different purposes. However, residual confounding and misclassification may have lead to an underestimate of the results, regarding the study setting (cross sectional study). Other limitations of our data are the lack of information of sun exposure and outdoors activities. Furthermore, longer time difference between the cohorts may have shown a stronger association of traditional marine diet and vitamin D concentration.

Public health relevance

Our observations indicate the challenges that lie ahead for pregnant women who aim to maintain adequate 25(OH)D levels by simply relying on eating traditional Faroese food. In the study of the elderly in the Faroe Islands, Dalgård et al concluded that higher supplements of Vitamin D and/or fortification might be necessary to maintain adequate 25(OH)D levels in order to prevent diseases and to ensure optimal body function and bone health. In Cohort 5, the majority of the women indicate that they take vitamin D supplements. However, the results suggest that this may not be enough to ascertain infants with sufficient levels. It could therefore be discussed whether the recommendations should be increased to pregnant women, especially as this is not just a Faroese problem, according to recent unpublished Danish studies. In addition, these recommendations may not take into consideration that there has been a general increase in BMI and a general decrease in marine food in recent years. Factors that may both influence maternal vitamin D concentration.

There should be further investigations on whether dietary supplements recommendations should be modified with regards to vitamin D or whether there should be made vitamin D enrichment in Faroese products.

Marine diets association to vitamin D status should be studied closer, with questionnaires specific for marine food rich in vitamin D and measurements of free vitamin D in blood of the participants. The seabird that is richest in vitamin D, the puffin, is threatened on the Faroe Islands, and therefore has been preserved for some years.
Future studies should look at outdoor activities, in relation to vitamin D. As Engelsen et al study suggests that sunlight in Faroe Islands is short on UVB radiation from October to February (7) it would be interesting to examine if it is possible to get UVB radiation, for the rest of the year in an amount high enough to raise the 25(OH)D level, by more frequent outdoors activity.

As previously described, vitamin D deficiency may increase the risk of different diseases such as cardio-vascular diseases and diabetes. Thus it is of great importance to aim for higher vitamin D levels in Faroese pregnant women. The challenge is big, especially because of the lack of UVB exposure from October to February. It could be relevant to enrich Faroese dairy products with vitamin D, and take a closer look at the recommendations in other countries of the same or higher latitude, such as Iceland and Finland. Additionally, it should be recommended to eat more oily fish such as mackerel, herring, halibut and salmon, the most recent government recommendations are to eat more fish, without preferences for oily types of fish. Furthermore it should be recommended to take a daily vitamin D supplement.

Conclusions

The lowered intake of traditional food in Faroese pregnant women between the two birth periods does not seem to explain the decreased concentration of 25(OH)D in newborns.

However, the decrease may be partly explained by a general increase in population BMI and some unidentified differences between the birth periods.

It is not possible, regarding our analysis, to give Pilot Whale the credit as a significant source of vitamin D for pregnant women in the two cohorts.

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Table 1. Participant's characteristic in Cohort 3, C3 (n = 570) and Cohort 5, C5 (n = 447)

	N (%)		Mean (SD)		P value
	C3	C5	C3	C5	
Women's age (years)			29.3 (5.2)	29.8 (5.5)	0.0729
15-24 Yr	111 (19.5)	73 (16.3)			0.011
25-29 Yr	159 (27.9)	139 (3.1)			
30-34 Yr	213 (37.4)	138 (30.9)			
35+ Yr	87 (15.3)	97 (21.7)			
Women's body mass index (kg/m^2)			23.8 (4.0)	24.3 (4.3)	0.0286
<18.5	20 (3.5)	11 (2.5)			0.238
18.5-24.9	381 (66.8)	281 (62.9)			
25-29.9	127 (22.3)	110 (24.6)			
≥30	42 (7.4)	45 (10.1)			
Women's weight gain during pregnancy			14.8 (5.1)	15.4 (5.6)	0.0408
	Parity				
primipara	149 (26.1)	129 (28.9)			0.289
nullipara	191 (33.5)	161 (36)			
secundipara	154 (27.0)	98 (21.9)			
tercipara	76 (13.3)	59 (13.2)			
Birth season[a]					
Spring	124 (21.8)	127 (28.4)			<0.001
Summer	153 (26.8)	73 (16.3)			
Autumn	136 (23.9)	130 (29.1)			
Winter	157 (27.5)	117 (26.2)			
Umbilical cord S-25OHD (nmol/l)			27.9 (18.2)	25.3 (18.8)	0.0127
<12.5	116 (20.4)	117 (26.2)			0.161
12.5 – 25	180 (31.6)	128 (28.6)			
25 – 50	210 (36.8)	159 (35.6)			
>50	64 (11.2)	43 (9.6)			
Alcohol consumption during pregnancy					
No	333 (58.4)	424 (94.9)			<0.001
yes	237 (41.6)	23 (.1)			
Smoking during pregnancy					
No	417 (73.2)	381 (85.2)			<0.001
yes	153 (26.8)	66 (14.8)			
Women's diabetic illness					
No	522 (91.6)	435 (97.3)			<0.001
DM 1	5 (0.9)	0			
GDM	43 (7.5)	12 (2.7)			
Women's educational level					
None/low	272 (47.7)	144 (32.2)			<0.001
Short	19 (3.3)	117 (26.2)			
Medium	135 (23.7)	135 (30.2)			
High	144 (25.3)	51 (11.4)			
Highest education of the parents[b]					
None/low	110 (19.3)	68 (15.2)			<0.001
Short	1 (0.2)	18 (4)			
Medium	153 (26.8)	127 (28.4)			
High	306 (53.7)	234 (52.3)			

Abbreviations: Umbilical cord S-25OHD, umbilical cord serum 25-hydroxy vitamin D; DM 1, diabetes mellitus type 1; GDM, gestational diabetes mellitus. [a]Birth season are for spring (March, April and May), summer (June, July and August), autumn (September, October and November) and winter (December, January and February).
[b]Highest education of the parents refers to the highest educational level of the women or her partner/father to be.

Table 2. Intake of traditional marine food[a] by cohort and vitamin D level.

	Vitamin D level in Cohort 3 (n = 267)			Vitamin D level in cohort 5 (n = 433)		
	<50 nmol/l n (%)	≥50 nmol/l n (%)	P value	<50 nmol/l n (%)	≥50 nmol/l n (%)	P value
Fish meals ≥2 pr. week						
No	63 (23.6)	11 (4.1)	0.467	167(38.6)	15 (3.5)	0.383
Yes	157 (58.8)	36 (13.5)		224 (51.7)	27 (6.2)	
Pilot whale meat[b]						
No	87 (32.6)	21 (7.9)	0.515	316 (73)	29 (6.7)	0.072
Yes	133 (49.8)	26 (9.7)		75 (17.3)	13 (3.0)	
Pilot whale blubber[b]						
No	134 (50.2)	32 (12)	0.357	371 (85.7)	37 (8.5)	0.073
Yes	86 (32.2)	15 (5.6)		20 (4.6)	5 (1.2)	
Seabirds[b]						
No	80 (30)	16 (6)	0.763	262 (60.5)	30 (6.9)	0.561
Yes	140 (52.4)	31 (11.6)		129 (29.8)	12 (2.8)	
Traditional marine food habit[c]						
No	182 (68.2)	39 (14.6)	0.967	385 (88.9)	39 (9.0)	0.015
Yes	38 (14.2)	8 (3)		6 (1.4)	3 (0.7)	

[a]All intake of traditional marine food items are of women's intake during pregnancy (yes/no). [b]Are intake of one or several meals during pregnancy. [c]Traditional marine food habit include all of the following: Fish meals ≥2 pr. week, Pilot whale meat, Pilot whale blubber and Seabirds (yes/no).

Table 3

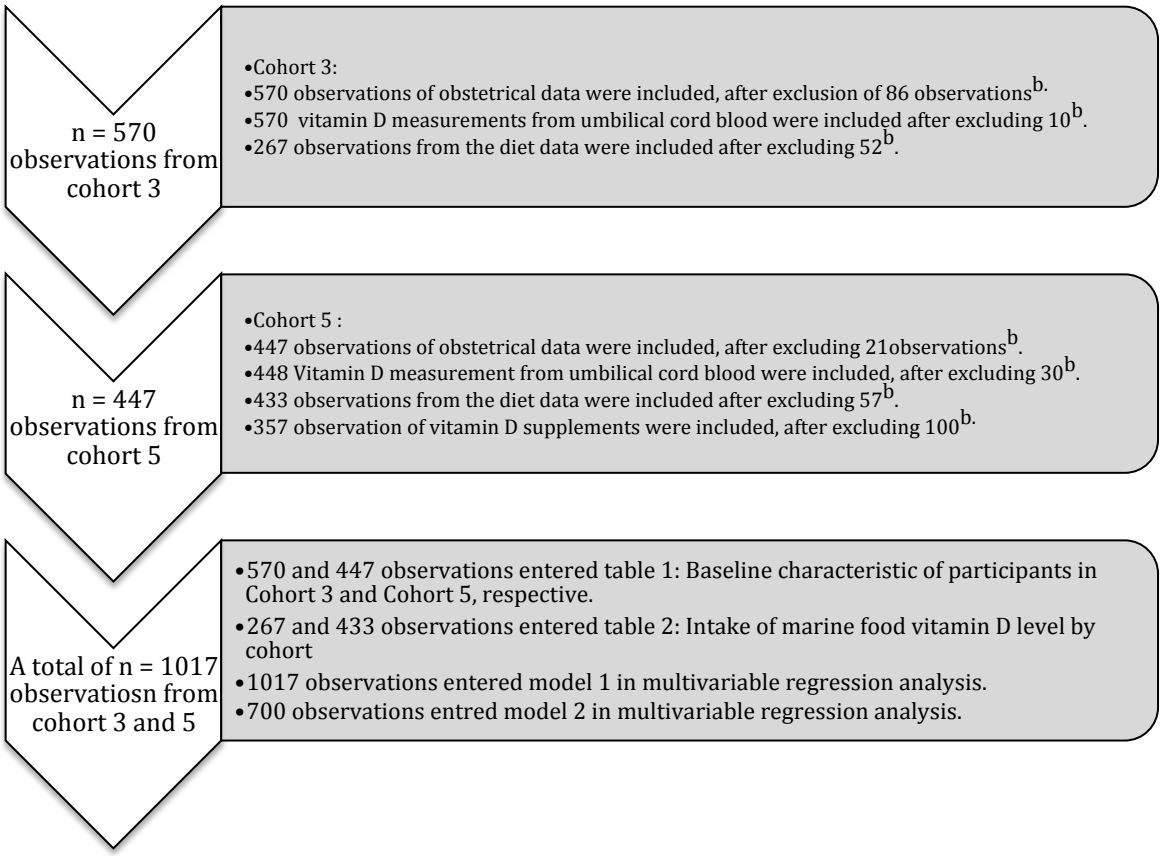
	Sample	Number	Cholecalciferol (vitamin D3), µg/100g	SD	Min – Max
Samples from the genital opening region	Pilot Whale blubber	15	1.04	1.27	0.25 – 4.42
	Pilot Whale meat	14	0.65	0.74	0.25 - 2.71
Samples collected for eating	Pilot Whale blubber	3	1.57	0.12	1.35 – 1.42
	Pilot Whale meat	1	0.25		
All samples included	Pilot Whale blubber	18	1.1	1.15	0.25 – 4.42
	Pilot Whale meat	15	0.63	0.72	0.25 – 2.71

Table 4. Multivariable regression analysis with umbilical cord S-25OHD as dependent variable

variable	Model 1[a] (n = 1017)		Model 2[b] (n = 700)	
	ß-estimate (95% CI)	P value	ß-estimate (95% CI)	P value
Parity				
Nullipara	Reference			
Primipara	-1-79 (-4.64; 1.06)	0.219	-3.62 (-7.25; -0.01)	0.051
Secundipara	-3.74 (-6.85; -0.63)	0.019	-7.87 (-11.86; -3.89)	<0.001
Tercipara+	-4.99 (-8.69; -1.30)	0.008	-5.68 (-10.54; -0.83)	0.022
BMI before pregnancy				
	-0.57 (-0.83; -0.31)	<0.001	-0.87 (-0.87; -0.23)	0.001
Diabetic condition				
no	Reference			
DM 1	5.10 (-10.46; 20.66)	0.520	7.27 (-13.71; 28.25)	0.497
GDM	-6.43 (-11.23; -1.57)	0.010	-7.35 (-13.75; -0.95)	0.025
Alcohol consumption				
	0.91 (-1.86; 3.69)	0.517	2.31 (-1.71; 6.33)	0.259
Smoking				
	-2.41 (-5.10; 0.29)	0.080	-2.23 (-5.67; 1.21)	0.205
Season of childbirth				
Spring	Reference			
Summer	8.34 (5.13; 11.55)	<0.001	9.89 (5.85; 13.94)	<0.001
Autumn	1.72 (-1.32; 478)	0.267	6.92 (3.05; 10.79)	<0.001
winter	-0.80 (-3.83; 2.23)	0.603	-0.63 (-4.34; 3.08)	0.737
Highest education of the parents[c]				
None or low	Reference			
Short	2.56 (-6.20; 10.64)	0.605	4.87 (-4.46; 14.01)	0.296
Medium	3.32 (-0.27; 6.57)	0.071	4.81 (0.47; 9.15)	0.030
Long	2.64 (-0.49; 5.78)	0.099	5.01 (1.01; 9.02)	0.014
Cohort 5	-2.64 (-2.05; 0.45)	0.211	-2.68 (-4.46; -0.89)	0.003
Fish meal ≥ 2 pr. week			2.06 (-0.85; 4.97)	0.165
Pilot whale meat			2.68 (1.00; 6.36)	0.154
Pilot whale blubber			-3.62 (-8.23; 0.98)	0.123
Seabirds			0.15(-2.75; 3.05)	0.919

Abbreviations: Umbilical cord S-25OHD, umbilical cord serum 25-hydroxy vitamin D; DM 1, diabetes mellitus type 1; GDM, gestational diabetes mellitus. [a]In model one smoking and alcohol consumption are for the women during pregnancy. [b]In model two the variables are the women's intake of marine diet during pregnancy. [c] Highest education of the parents refers to the highest educational level of the women or her partner/father to be.

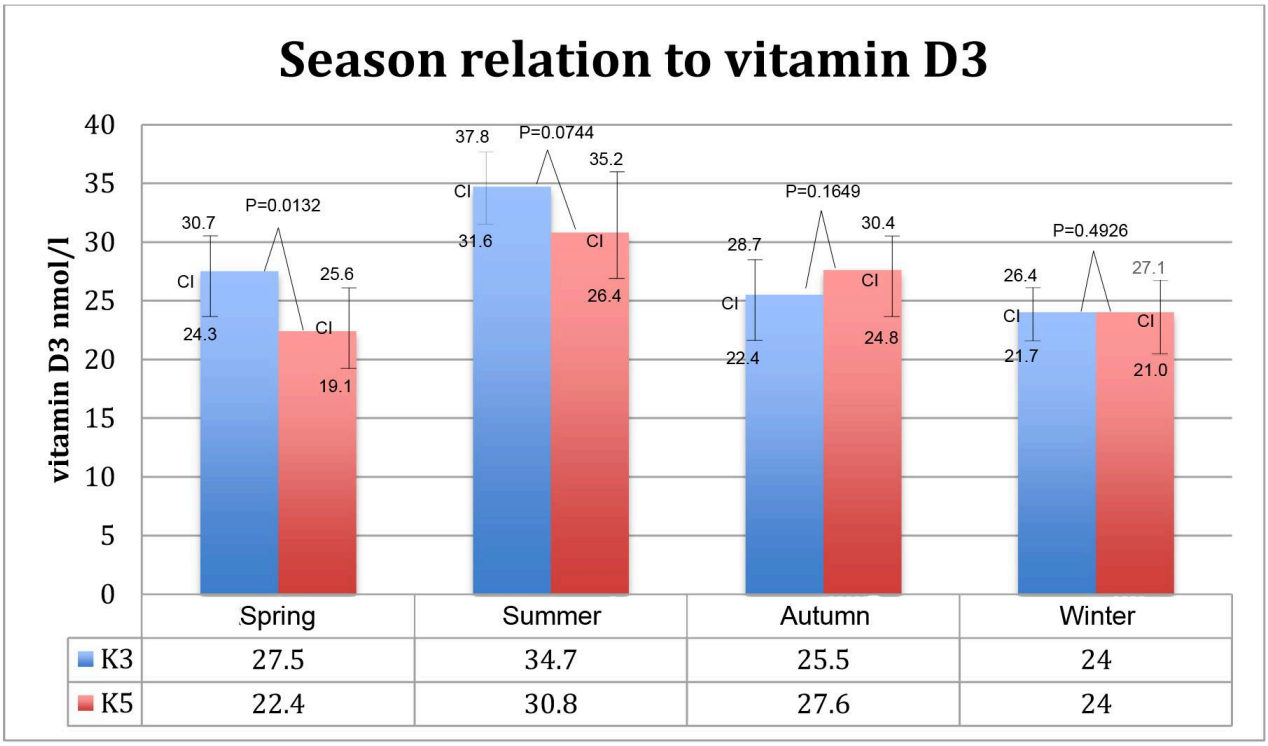
Fig 1: Flow chart over included observation from CHEF [a] research project



[a]CHEF: Children's Health and the Environment in the Faroese

[b]All observation not containing full information regarding vitamin D level and obstetrical and diet data were excluded.

Figure 2.


Season relation to vitamin D3
vitamin D3 nmol/l
40
35
30
25
20
15
10
5
0
P=0.0132
30.7
CI
24.3
25.6
CI
19.1
37.8
P=0.0744
CI
31.6
35.2
CI
26.4
P=0.1649
28.7
CI
22.4
30.4
CI
24.8
P=0.4926
26.4
CI
21.7
27.1
CI
21.0
Spring
Summer
Autumn
Winter
K3
27.5
34.7
25.5
24
K5
22.4
30.8
27.6
24